

WE CLAIM:

1. An oligonucleotide of about 20 to about 40 nucleotides that hybridizes specifically to a sequence contained in a *B. anthracis* target sequence consisting of SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25, SEQ ID NO:26, SEQ ID NO:34, a complementary sequence, or RNA equivalent of any one of the target sequences.
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2. An oligonucleotide of claim 1 that hybridizes specifically to a *pagA* target sequence contained in the sequence consisting of SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, or SEQ ID NO:24, a complementary sequence, or RNA equivalent of any one of the *pagA* target sequences.
- 10 3. An oligonucleotide of claim 2 that hybridizes specifically to the *pagA* target sequence contained in SEQ ID NO:21, wherein the oligonucleotide has a sequence consisting of SEQ ID NO:1 or SEQ ID NO:2.
4. An oligonucleotide of claim 2 that hybridizes specifically to the *pagA* target sequence contained in SEQ ID NO:22, wherein the oligonucleotide has a sequence consisting of SEQ ID
15 NO:3 or SEQ ID NO:4.
5. An oligonucleotide of claim 2 that hybridizes specifically to the *pagA* target sequence contained in SEQ ID NO:23, wherein the oligonucleotide has a sequence consisting of SEQ ID NO:5 or SEQ ID NO:6.
6. An oligonucleotide of claim 2 that hybridizes specifically to the *pagA* target sequence
20 contained in SEQ ID NO:24, wherein the oligonucleotide has a sequence consisting of SEQ ID NO:7 or SEQ ID NO:8.
7. An oligonucleotide of claim 1 that hybridizes specifically to a *capB* target sequence contained in the sequence consisting of SEQ ID NO:25 or SEQ ID NO:26, a complementary sequence, or RNA equivalent of any one of the *capB* target sequences.
- 25 8. An oligonucleotide of claim 7 that hybridizes specifically to the *capB* target sequence contained in SEQ ID NO:25, wherein the oligonucleotide has a sequence consisting of SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, or SEQ ID NO:12.
9. An oligonucleotide of claim 7 that hybridizes specifically to the *capB* target sequence contained in SEQ ID NO:26, wherein the oligonucleotide has a sequence consisting of SEQ ID
30 NO:13 or SEQ ID NO:14.

10. An oligonucleotide of about 18 to 40 bases that hybridizes specifically to a 16S rRNA or DNA encoding a 16S rRNA sequence of a *Bacillus* species contained in a target sequence consisting of SEQ ID NO:31, a complementary sequence, or RNA equivalent thereof.
11. An oligonucleotide of claim 10, wherein the oligonucleotide has a sequence consisting of SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:37, SEQ ID NO:38, or SEQ ID NO:39.
12. An oligonucleotide of about 20 to 50 bases that hybridizes specifically to a 23S rRNA or DNA encoding a 23S rRNA sequence of a *Bacillus* species contained in a target sequence consisting of SEQ ID NO:32, a complementary sequence, or RNA equivalent thereof.
13. An oligonucleotide of claim 12, wherein the oligonucleotide has a sequence consisting of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:35, or SEQ ID NO:36.
14. An oligonucleotide of claim 1, wherein the oligonucleotide has a DNA or RNA backbone, or mixed DNA and RNA backbone, or contains at least one 2'-methoxy RNA group linking the bases.
15. An oligonucleotide of claim 10, wherein the oligonucleotide has a DNA or RNA backbone, or mixed DNA and RNA backbone, or contains at least one 2'-methoxy RNA group linking the bases.
16. An oligonucleotide of claim 12, wherein the oligonucleotide has a DNA or RNA backbone, or mixed DNA and RNA backbone, or contains at least one 2'-methoxy RNA group linking the bases.
17. An oligonucleotide of claim 1, wherein the oligonucleotide has a signal-producing label linked directly or indirectly to the oligonucleotide.
18. An oligonucleotide of claim 10, wherein the oligonucleotide has a signal-producing label linked directly or indirectly to the oligonucleotide.
19. An oligonucleotide of claim 12, wherein the oligonucleotide has a signal-producing label linked directly or indirectly to the oligonucleotide.
20. A method of detecting *B. anthracis* nucleic acid in a sample comprising the steps of:
providing a sample containing *B. anthracis* nucleic acids;
providing at least one probe that hybridizes specifically to a *pagA* target sequence contained in a pXO1 plasmid and at least one probe that hybridizes specifically to a *capB* target

sequence contained in a pXO2 plasmid;

hybridizing specifically at least one probe to the *pagA* target sequence, or at least one probe to the *capB* target sequence, or at least one probe to the *pagA* target sequence and at least one probe to the *capB* target sequence; and

5 detecting the presence of at least one probe hybridized to the *pagA* target sequence or to the *capB* target sequence to indicate the presence of *B. anthracis* in the sample.

21. The method of claim 20, wherein the *pagA* target sequence is contained in the sequence of SEQ ID NO:33, or a complementary sequence, or RNA equivalent thereof, and wherein the *capB* target sequence is contained in the sequence of SEQ ID NO:34, or a complementary
10 sequence, or RNA equivalent thereof.

22. The method of claim 20, wherein the *pagA* target sequence is contained in a sequence consisting of SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, or SEQ ID NO:24, a complementary sequence, or RNA equivalent of any one of these sequences, and the *capB* target sequence is contained in a sequence consisting of SEQ ID NO:25 or SEQ ID NO:26, a complementary
15 sequence, or RNA equivalent of any one of these sequences.

23. The method of claim 20, wherein the hybridizing step includes at least one probe specific for the *pagA* target sequence which is an oligonucleotide having a sequence of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, or SEQ ID NO:8.

20 24. The method of claim 20, wherein the hybridizing step includes at least one probe specific for a *capB* target sequence which is an oligonucleotide having a sequence consisting of SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:13, or SEQ ID NO:14.

25 25. The method of claim 20, further comprising the steps of providing at least one probe that hybridizes specifically to a 16S or 23S rRNA sequence or DNA encoding a 16S or 23S rRNA sequence conserved among species of the *B. cereus* complex, hybridizing the at least one probe to the 16S or 23S rRNA sequence or DNA encoding the 16S or 23S rRNA sequence conserved among species of the *B. cereus* complex, and detecting the presence of at least one probe hybridized to the 16S or 23S rRNA sequence or DNA encoding the 16S or 23S rRNA sequence conserved among species of the *B. cereus* complex, thereby indicating the
30 presence of a *B. cereus* complex organism in the sample.

26. The method of claim 25, wherein the at least one probe that hybridizes specifically to a 16S rRNA or DNA encoding a 16S rRNA sequence is an oligonucleotide of 18 to 40 bases that hybridizes specifically to a sequence contained in the sequence consisting of SEQ ID NO:31, a complementary sequence, or RNA equivalent thereof.

5 27. The of claim 26, wherein the oligonucleotide has a sequence consisting of SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:37, SEQ ID NO:38, or SEQ ID NO:39.

28. The method of claim 25, wherein the at least one probe that hybridizes specifically to a 23S rRNA or DNA encoding a 23S rRNA sequence is an oligonucleotide of 20 to 50 bases that
10 hybridizes specifically to a sequence contained in the sequence consisting of SEQ ID NO:32, a complementary sequence, or RNA equivalent thereof.

29. The method of claim 28, wherein the oligonucleotide has a sequence consisting of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:35, or SEQ ID NO:36.

30. The method of claim 25, wherein the providing step further includes providing a probe
15 that hybridizes specifically to a genetic sequence present in eubacterial species, wherein the hybridizing step further includes hybridizing the probe specifically to the genetic sequence present in eubacterial species, and wherein the detecting step further includes detecting the probe hybridized to the genetic sequence present in eubacterial species, thereby indicating that the method steps have been performed properly when no *Bacillus* sequences are detected
20 in the assay.

31. The method of claim 30, wherein the probe has a sequence consisting of SEQ ID NO:40, and wherein detecting the probe of SEQ ID NO:40 indicates the presence of a eubacterium in the sample.

32. A kit for practicing the method of claim 20, comprising at least one probe that hybridizes
25 to a sequence contained in the *pagA* target sequence consisting of SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, or SEQ ID NO:24, a complementary sequence, or RNA equivalent of any one of these sequences, and at least one probe that hybridizes specifically to a sequence contained in the *capB* target sequence consisting of SEQ ID NO:25 or SEQ ID NO:26, a complementary sequence, or RNA equivalent or any one of these sequences.

33. The kit of claim 32, comprising at least one probe specific for the *pagA* target sequence which is an oligonucleotide having a sequence of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, or SEQ ID NO:8, and at least one probe specific for a *capB* target sequence which is an oligonucleotide having a sequence consisting of SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:13, or SEQ ID NO:14.